REMARKS/ARGUMENTS

The present amendment is submitted in accordance with the *Revised Amendment Format* as set forth in the Notice provided on the USPTO web site for the Office of Patent Legal Administration; Pre-OG Notices; signed 1/31/03.

Claims 1-21 are pending in the application. No claims are allowed. Claims 22-23 have been canceled without prejudice to subsequent revival. Applicants reserve the right to prosecute claims 22-23 in a divisional application.

Claims 1, 3, 4, 9, 10, 12, 14 and 20 have been amended. Entry of the amendment, reconsideration of the rejection, and allowance of claims 1-21 are requested.

The Amendment

In order to expedite prosecution of the application and advance the case toward allowance, the claims have been amended. No new matter was introduced by this amendment.

Claims 1 and 12 have been amended to clarify that the method includes a step of incubating the serum free cell culture of VERO cells infected with HAV to propagate the HAV "at reduced temperature". Support for this amendment can be found on page 6, paragraph [018], lines 5-6.

Claims 1 and 12 have also been amended to clarify that infected cells release at least 50% of viral antigen into the cell supernatant; and that it is the HAV released into the supernatant of the cell culture medium that is being harvested. Support for this amendment can be found on page 12 of the specification, paragraph [041], lines 8-9.

Claims 3 and 14 have been amended to correct the claim dependency and to provide for proper antecedent basis.

Claims 4 and 15 have been amended to correct for proper Markush group language as requested by the Examiner.

Claim 9 has been amended to add the term "purified" before "trypsin-like enzyme". Support for this amendment can be found on page 9 of the specification, paragraph [028], line 3.

Claims 10 and 20 have been amended to clarify that it is the cells bound to the microcarrier that continuously produce and release HAV into the cell culture medium for at least 60 days. Support for this amendment can be found on page 10 of the specification, paragraph [033].

Rejection Under 35 U.S.C. §112

Claims 3-5, 10, 15-16, and 20 have been rejected under 35 U.S.C. §112, second paragraph for being allegedly indefinite.

The office action indicates that claim 3 shows insufficient antecedent basis for the claim limitation "said temperature". Claim 3 has been amended to correct the claim dependency and now depends on claim 2. Claim 2 refers to a temperature of about 37°C, thus, there is now proper antecedent basis for the term "said temperature" in claim 3.

Claims 4 and 15 are indicated to recite improper Markush group language and the claims have been corrected accordingly.

In claims 10 and 20, the term "HAV is continuously produced for at least 60 days" is allegedly unclear. The Examiner requires clarification to whether one culture produces for 60 days or a fresh culture replaces an older one and continues production. Claims 10 and 20 have been amended to clarify that it is the cells bound to the microcarrier that continuously produce and release HAV into the cell culture medium for at least 60 days. As the specification describes on page 7, paragraph [022], the instant invention provides production of HAV, wherein HAV is continuously released into the cell culture supernatant; while there are no prior art methods that continuously produce HAV over such a long period of time. The specification also elaborates on the procedure, wherein a microcarrier culture system and cell culture perfusion are used. The medium containing the virus is continuously removed from the cell culture and fresh culture medium is added and continuously perfused (see page 7, paragraph [022], lines 6-8). In light of the specification, amended claims 10 and 20 are clear and definite. Thus, the skilled artisan would have no difficulty discerning the meaning of "the cells bound to the microcarrier continuously produce and release HAV into the cell culture medium for at least 60 days" in light of the specification.

Rejection Under 35 U.S.C. §102

Claims 1, 2, 4, 5, 8, 9, 11-13, 15, 16, 18 and 19 are rejected under 35 U.S.C. §102(b) as being allegedly anticipated by Kistner *et al.* (WO 96/15231).

The Office Action states that Kistner teaches a method of cultivating, harvesting and purifying influenza virus that can be applied to HAV and that Kistner's method produces HAV which remains in the cells and is released into the supernatant.

The rejection is respectfully traversed to the extent that the rejection applies to the claims as amended.

"In order for a rejection under §102(b) to be valid, each and every element of the claim must be found in the prior art reference." (MPEP 2131; *In re Royka and Martin,* 180 USPQ 580 (CCPA 1974)).

The amended claims are directed to a method of producing HAV by providing a serum free cell culture of VERO cells bound to a microcarrier, wherein the HAV infected cells are propagated at reduced temperature and release at least 50% of viral antigen into the cell supernatant. The viral antigen is then harvested from the supernatant. As stated in the specification on page 7, paragraph [021], the Applicants found that HAV is continuously released into the cell culture medium supernatant which was unexpected, because prior art using VERO cells as a host for HAV disclosed that ***HAV could only be found intracellulary*** and virus produced had to be obtained from the cells (U.S. 4,783,407).

In their examples, the Applicants disclose how VERO cells cultured at 34°C in serum free medium continuously release viral antigen into the cell culture supernatant and about 50% of the viral antigen is found in the supernatant of the culture medium. This is contrasted to the same method with serum-containing medium wherein viral antigen is dominantly found in the cell pellet like in most prior art methods (see page 12, paragraph [040]). Tables 1 and 2 (page 13) further illustrate how increasing numbers of viral antigen are deposited into the supernatant with higher cell passages. On page 14, paragraph [044], the Applicants show that virus titers of 50,000,000 and 200,000,000 (per 5,000,000 cells) are obtained in the cell pellet and cell culture supernatant, respectively, which demonstrates that viral antigen is continuously released into the cell culture supernatant by serum-free VERO cells. Thus, three weeks post

infection, ***the percentage of viral antigen in the supernatant is about 50%*** (see Table 2). During long-term propagation of microcarrier bound serum free VERO cells, the virus containing supernatant is collected and stored at 4°C when antigen is detected in the medium. The HAV antigen is then further purified from the cell culture supernatant.

In comparison, Kistner *et al.* (Kistner) describe a method of producing influenza virus by utilizing vertebrate cells cultured under protein-free conditions. The Examiner points to page 31, lines 8-10, wherein Kistner describes that not all virus particles produced by the cells are released into the supernatant; and that these particles are still associated with the cells of the cellular biomass. In order to obtain an increased virus yield, Kistner harvests the virus particles from the supernatant *as well as* the virus associated with the cellular biomass. In fact, on page 31, lines 17-18, Kistner indicates that the virus found in the cells of the cellular biomass is released from the cells by lysis, wherein various cell lysis methods are disclosed. Hence, Kistner does not disclose a method wherein HAV infected VERO cells release at least 50% of viral antigen into the cell supernatant. Rather, Kistner discloses a method that requires releasing intracellular virus from the cells of the cellular biomass as indicated on page 32, lines 7-8. Moreover, Kistner relies on the virus derived from the cellular biomass to obtain maximum virus and virus antigen yield (see page 32, lines 16-24); and Kistner does not disclose a method wherein HAV infected cells are propagated at reduced temperature. As a result, Kistner does not anticipate the instant invention.

In light of the amendment and arguments presented above, Applicants respectfully request that the rejection of claims 1, 2, 4, 5, 8, 9, 11-13, 15, 16, 18 and 19 under 35 U.S.C. §102(b), be withdrawn.

Rejection Under 35 U.S.C. §103

Claims 3, 6, 7, 14, 17 and 21 are rejected under 35 U.S.C. §103(a) as being allegedly unpatentable over Kistner *et al.* (WO 96/15231) as applied to claims 1, 2, 4, 5, 8, 9, 11-13, 15, 16, 18 and 19 above, and further in view of Purcell *et al.* (U.S. Patent No. 4,894,228), Leu *et al.* (WO 95/24468), Pellegrini *et al.* (U.S. Patent No. 5,607,851) and Shih *et al.* (US Patent No. 5,980,901).

The Office Action alleges that one would have had a reasonable expectation of success that the HAV strain HM175/7 would have grown in Kistner's cell culture method because Purcell teaches that the preferred method of propagating HM175/7 is in cell culture. The Office Action further alleges that one would have been motivated to infect the cells of Kistner with a m.o.i. of 0.05 to 1, as is standard practice, as evidenced by Leu. It is further asserted that one would have had a reasonable expectation of success that isopycnic centrifugation would have separated the particles of Kistner from the media because Shih separates particles from the media using this method.

The rejection is respectfully traversed to the extent that the rejection applies to the claims as amended.

As the Examiner is aware, the prior art references must teach or suggest all the limitations of the claims. *In re Wilson*, 165 U.S.P.Q. 494, 496 (C.C.P.A. 1970). However, in this case the prior art references do not teach or suggest all the limitations of the claims and therefore, the obviousness rejection should be withdrawn.

As discussed above, Kistner does not teach a method wherein HAV infected VERO cells are propagated at reduced temperature and release at least 50% of viral antigen into the cell supernatant. The other four references cited by the Examiner do not discuss this limitation either. In fact, the Examiner has pointed to nothing in the secondary references that teaches or suggests this element of the claimed method. Since in each case, all of the claim limitations are not suggested by the combination of references, the rejection should be withdrawn.

As the Examiner is further aware, obviousness can only be established by combining or modifying the teachings of the prior art to produce the claimed invention where there is some teaching, suggestion, or motivation to do so found either in the references themselves or in the knowledge generally available to one of ordinary skill in the art. *In re Fine*, 837 F.2d 1071, 5 USPQ2d 1596 (Fed. Cir. 1988); *In re Jones*, 958 F.2d 347, 21 USPQ2d 1941 (Fed. Cir. 1992).

As shown above, Kistner does not anticipate the instant invention because Kistner does not teach a method wherein HAV infected VERO cells are propagated at reduced

temperature and release at least 50% of viral antigen into the cell supernatant. Thus, Kistner's disclosure coupled with the knowledge of an attenuated hepatitis A virus genome and mutations that are involved in cell culture adaptation (see Purcell) and/or the knowledge that a hepatitis A virus can be grown in MRC-5 cells, wherein cells are lysed with a detergent buffer system (see Leu, page 8, line 6) does not teach the skilled artisan to produce the claimed invention. Further adding the knowledge that MRC-5 cells can be infected with hepatitis A virus with a m.o.i. of 0.05 and purified by lysing the cells with detergent (see Pellegrini, see claims) and/or the knowledge that a defective interfering particle of a hepadnavirus can be purified via isopycnic centrifugation (see Shih) still does not teach the skilled artisan how to practice the claimed invention. There is simply no motivation to combine the references because applying Kistner's method to any or all of the methods taught by Purcell, Pellegrini, Leu and/or Shih would not lead to the claimed method.

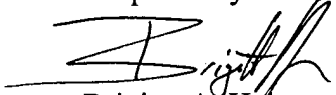
In light of the amendment an arguments presented above, Applicants respectfully request that the rejection of claims 3, 6, 7, 14, 17 and 21 under 35 U.S.C. §103(a), be withdrawn.

CONCLUSION

In view of the foregoing, Applicants believe all claims now pending in this Application are in condition for allowance. The issuance of a formal Notice of Allowance at an early date is respectfully requested.

If the Examiner believes a telephone conference would expedite prosecution of this application, please telephone the undersigned at 650-326-2400.

Respectfully submitted,



Brigitte A. Hajos
Reg. No. 50,971

TOWNSEND and TOWNSEND and CREW LLP
Two Embarcadero Center, Eighth Floor
San Francisco, California 94111-3834
Tel: 650-326-2400
Fax: 415-576-0300
Attachments

60078476 v1